

Measuring Vitamin A in Sugar

1. Method Principle and Application

[iCheck Fluoro](#) is a portable fluorometer for quantitative determination of the vitamin A content in vitamin premixes, sugar, flour and milk. The method is based on the fluorescence of the retinol molecule (excitation at 340 nm, emission at ≥ 400 nm). The device algorithm calculates vitamin A concentration in μg retinol equivalents (RE) per liter ($\mu\text{g RE/L}$). The measurement range of iCheck Fluoro is 50 – 3000 $\mu\text{g RE/L}$.



2. Vitamin A in Fortified Sugar

The expected vitamin A concentration in fortified sugar is generally 10 – 30 mg RE/kg. The most common form of vitamin A added to sugar is retinyl palmitate. Other esterified forms, such as retinyl acetate, are less frequently used.

3. iCheck Fluoro Performance with Sugar

iCheck Fluoro performance is assessed following a standardized process that combines assessment of precision, trueness and a comparison to a reference method (i.e., high-performance liquid chromatography - HPLC). The detailed description of this process is provided in the [iCheck Fluoro Performance Guide](#).

Performance of iCheck Fluoro with sugar and sugar preblend has been assessed in internal validations. Below is a table detailing the observed precision and recovery.

Table 1. iCheck Fluoro Performance with Sugar and Sugar Preblend

Sample Type	Fortificant	Added Vitamin A Conc.	iCheck Fluoro performance
Fortified sugar	Retinyl palmitate	5 – 15 - 30 mg/kg	Recovery: 119 \pm 4%
White sugar preblend		Range: 13,500 – 19,500 mg/kg	Measured: 16,407 \pm 217 mg/kg
Brown sugar preblend			Measured: 13,114 \pm 95 mg/kg

Additionally, an independent study was conducted to compare the performance of iCheck Fluoro to HPLC. The results of this study are summarized in the table 2 [1].

Table 2. iCheck Fluoro Performance compared to HPLC [1]

Sample Type	Fortificant	Coefficient of variation iCheck Fluoro	Coefficient of variation HPLC	Correlation Coeff. HPLC vs. iCheck Fluoro (R²)
Sugar	Retinyl palmitate	Max.±10%	Max.±11%	>0.91

4. Analyzing Sugar Containing Vitamin A

The measurement range of iCheck Fluoro is 50-3000 µg RE/L. The sugar sample must be diluted in bottled or distilled water to fit the measurement range.

Weigh in the sugar according to Table 3 and record the exact weight.

Table 3. Dilution of Sugar for Vitamin A Quantification with iCheck Fluoro

Sample Type	Expected Vit. A Conc. [mg/kg]	Expected Vit.A Conc. [IU/kg]	Dilution	Sugar Sample Weight [g]	Volume of Water [mL]	Expected diluted sugar concentration [µg RE/L]
Sugar	10 - 30	30,000-100,000	1 : 20	25	500	500 – 1,500

- Transfer your sample to a bottle and dilute it to a total volume of 500 mL. Shake until fully solubilized.
- Do NOT use refrigerated water. Water must be brought to room temperature.
- Record the exact weight and total volume of your sample solution for dilution factor calculation.
- **Vitamin A is not stable in solution with water!** Proceed with measurement immediately.
- Vitamin A is not soluble but only dispersible in water. Therefore, if the diluted sugar is standing still the vitamin A will separate from the water. Shake the solution and immediately take it up into the syringe.
- Inject 0.5 mL of the sample solution into a new iCheck Fluoro reagent vial. Shake the vial vigorously for 10 seconds. Proceed as described in the [iCheck Fluoro User Manual](#).



5. Dilution Factor (DF) Calculation

The value displayed on iCheck Fluoro after measurement will reflect the concentration of vitamin A in the diluted sample. To obtain the original sugar vitamin A concentration, you must first calculate the dilution factor according to the following formula:

$$DF = \frac{\text{Total sample solution volume (mL)}}{\text{Sample weight (g)}}$$

Once you have calculated the dilution factor, multiply the iCheck Fluoro result by the dilution factor.

$$\text{Vitamin A in sugar} \left(\frac{\text{mg RE}}{\text{kg}} \right) = \text{iCheck Fluoro reading} \left(\frac{\mu\text{g RE}}{\text{L}} \right) \times \frac{DF}{1000}$$

6. Vitamin A Unit Conversion

Below, you can find the relationship between retinol equivalents and other units used for vitamin A measurement, and for converting retinol palmitate/acetate to retinol.

- **1 mg Vitamin A = 1 mg retinol = 1 mg RE**
- **1 mg RE = 3333 International Units (IU)**
- **0.3 IU = 0.001 mg RE = 1 g RE**
- **1 µg RE = 3.33 IU**
- **1 µg retinyl palmitate = 0.55 µg retinol**
- **1 µg retinyl acetate = 0.66 µg retinol**
- **1 mg = 1000 µg**

[1] Laillou A, Renaud C, Berger J, Moench-Pfanner R, Fontan L, Avallone S. Assessment of a portable device to quantify vitamin A in fortified foods (flour, sugar, and milk) for quality control. Food Nutr Bull. 2014.

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